BACTERIOLOGICAL STUDIES OF THE
PENETANGUISHENE – WAUBAUSHENE
AREA OF GEORGIAN BAY
1973,1974 AND 1976

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A BACTERIOLOGICAL STUDY OF THE PENETANGUISHENE-WAUBAUSHENE AREA OF GEORGIAN BAY, 1973 AND 1974.

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ABSTRACT

Waubaushene area during 1973 and 1974. The bacterial parameters were Total and Fecal Coliforms, Fecal Streptococci, Heterotrophic Bacterial Count and Pseudomonas aeruginosa. The results of both years indicated a trend to increased levels during the summer period but the bacterial levels and their geographical heterogeneity differed. The 1974 spring and summer results indicated somewhat poorer water quality than in 1973. The 1974 summer levels for Total Coliforms exceeded Ministry of the Environment recreational use criteria, however, they were significantly lower than levels obtained during an earlier survey in 1969. A significant cause of heterogeneity within a given survey was the elevated bacterial levels in the vicinity of the area's Sewage Treatment Plants in particular those at Penetang and Midland. A biweekly sampling during 1973 demonstrated the same seasonal trends but proved to be less sensitive to geographical differences in water quality and in general produced significantly higher Total Coliform results.

A Bacteriological Study of the Penetanguishene Waubaushene Area of Georgian Bay

Introduction

Surveys of the Penetanguishene-Waubaushene area were conducted in 1973 and 1974 as part of a continuing Ministry of the Environment program to monitor water quality and as part of the International Joint Commission - Upper Great Lakes Surveys. The surveys were designed to determine the water quality of the area, the influence of sewage treatment plant (STP) effluents and to provide data to monitor the effect of improvements planned for waste treatment in the area. An evaluation of survey design was also included in 1973.

The survey area (Fig. 1) is located eighty miles north-nortwest of Toronto along the southern extremity of the Canadian Shield. With the exception of Port Severn on the Severn River, the centres of population (Penetanguishene, Midland, Port McNicoll, Victoria Harbour and Waubaushene) have developed on the four bays (Penetang, Midland, Hog and Sturgeon) along the southern shoreline of the survey areas (Fig. 2). The survey stations were concentrated within these four bays.

Water Quality Surveys carried out by the O.W.R.C. (now the Ministry of the Environment) of Penetang Bay in 1964 and 1967, of Midland Bay in 1966 and of the entire area in 1969 indicated that some problems with water quality existed particularly in Penetang and Midland Bays (Jones 1970; Veal and Michalski, 1971).

At the time of the 1973 survey the waste treatment facilities were the same as in 1969. However, plans for the addition of phosphate removal to the existing STP's at Penetang, the Ontario Hospital (Penetang), and Midland were to be implemented. The Penetang and Ontario Hospital STP's were also to be expanded. The projects at the Ontario Hospital and Midland STP's were completed



FIGURE 1 - LOCATION OF THE SURVEY AREA IN THE GREAT LAKES

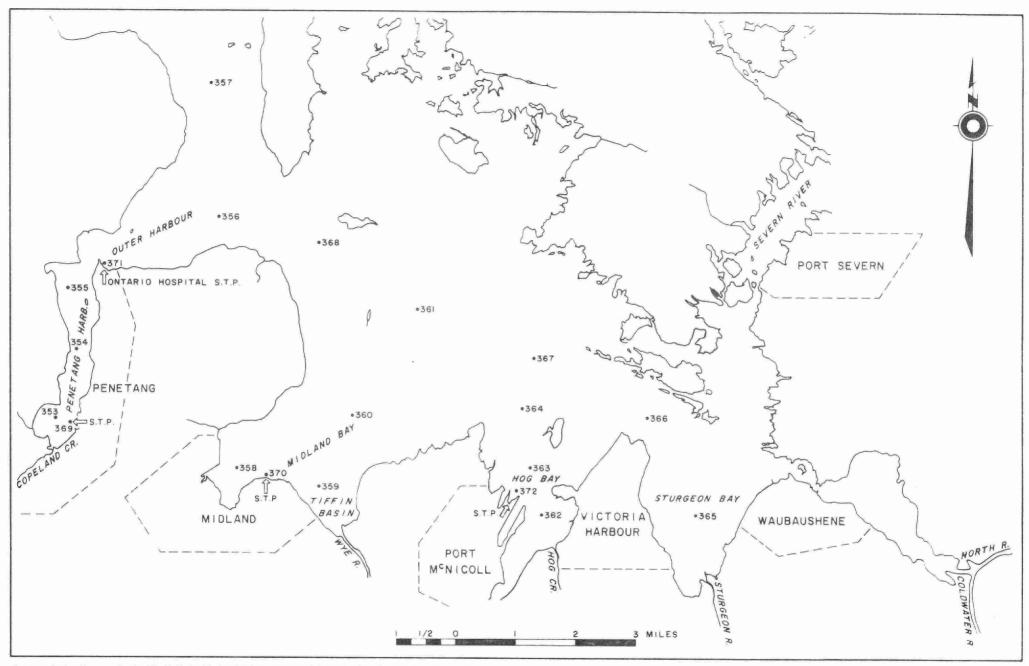


FIGURE 2 - PENETANGUISHENE-WAUBAUSHENE SURVEY AREA

early in 1974 although phosphate removal at the Midland Plant was still intermittent. The work at the Penetang Plant was to be completed in 1975. In addition to the above improvements, Port McNicoll was to convert from septic tank-tile systems to a sewage collector system combined with a secondary treatment plant. A few homes were connected to the system in November 1971 to permit operation throughout the winter and the system has continued to expand. Phosphate treatment will be initiated at the Port McNicoll plant if phosphate levels reach 1 ppm in the effluent. Victoria Harbour and Waubaushene remained on individual septic tank systems (Marton, 1975).

Methods

1. Field Procedures

1973

Bacteriological samples were collected at 20 stations, including the eleven used in the 1969 survey. The survey consisted of two parts: first, a biweekly sampling schedule from May 24, 1973 to September 12, 1973; second, three sevenday intensive surveys from May 16 to 22, July 9 to 14, and August 30 to September 5, 1973.

Samples were collected in sterile, autoclavable, polycarbonate, 250 ml bottles at a depth of approximately 1.5 m below the surface. The samples were immediately iced and shipped either to a mobile laboratory located in Penetanguishene (May and August surveys) or to the Toronto laboratory (biweekly samples and July survey).

1974

Two 5-day intensive surveys were carried out from July 23 to 27 and September 11 to 17. The same station locations were used as in 1973. In addition, a 3-day survey of the area was conducted during the period May 30 to June 6 as part of the Georgian Bay Nearshore survey.

The samples were analyzed in a mobile laboratory located in Penetanguishene.

2. Laboratory Procedures

All samples were analyzed for Total Coliform (TC), Fecal Coliform (FC) and Fecal Streptococci (FS). Membrane filtration analyses were conducted according to Standard Methods (13th edition) using m-Endo Agar Les (Difco) for TC, MacConkey Membrane Broth (Oxoid) for FC, and m-Enterococcus Agar (Difco) for FS. In 1974, all samples were also analyzed for Heterotrophic Bacteria (HB) and Pseudomonas aeruginosa (P. aer.).

The HB method consisted of a spot plate technique (Bousfield, Smith and Truemann, 1974) on Foot and Taylor Agar (Foot and Taylor, 1949) at 20°C for seven days. The P. aer. method utilized was the membrane filtration procedure of Levin and Cabelli (mPA). The incubation was at 41.5°C for 48 hours (Levin and Cabelli, 1972).

Analysis of the 1973 biweekly samples was conducted within 24 hours of sampling while the analysis of the samples for all other surveys was carried out within 12 hours.

3. Statistical Methods

Fluctuations in bacterial concentrations due to changing environmental conditions require that a great number of samples be taken to arrive at a mean value which is representative of a specific sample location or sampling area. The most appropriate mean for bacterial levels and this type of data is the geometric mean. The large amounts of bacteriological data generated from this survey necessitated statistical methods to summarize the results concisely and to facilitate an unbiased interpretation.

For the intensive surveys the daily results for each parameter were organized as replicate results for each station. The log geometric mean, the variance, and the standard error were then calculated for each parameter at each station.

The 1973 biweekly survey results were organized into four-time series' groups, May 24 to June 21, June 21 to July 31, July 31 to September 12, and September 12 to October 24. Each of these groups was then treated in the same way as the intensive surveys to obtain the group statistics for each parameter at each station.

Once the station group statistics had been obtained, an analysis of variance program (ANOVA) was used to group the stations into areas within the same statistical bacterial level. The ANOVA analysis was first performed on all survey stations. If the calculated F-ratio was less than the critical F-ratio (0.05 level), the stations were considered statistically the same and were summarized as a group with one set of overall group statistics. At the same time as the ANOVA analyses were performed, the homogeneity of the variances was also checked using Bartlett's X² test of homogeneity. If either the F or X² was significant, indicating a non similar grouping, stations that were judged to be significantly different,

based on a statistical circumspection of the data, were tested and, if necessary, eliminated until both the F-ratio and X² were nonsignificant. The withdrawn stations were regrouped with respect to geographic proximity. The calculations on all groups were repeated using the ANOVA program until each discrete group was homogeneous. The Student-t test (using the log GM and S.E.) was used to compare overlapping homogeneous areas between each of the three intensive surveys and between the four biweekly groupings.

It should be noted that because of the long time interval between sampling and the generally low levels of data for the biweekly groupings, one can be much more confident in the results obtained from the intensive surveys where these problems are not encountered. The biweekly data is useful in indicating overall trends but not as sensitive in defining specific problems.

Summary tables of the analysis of variance grouping of stations and the tests of significance between analysis of variance groups and/or stations are on file at the Great Lakes Laboratory, Microbiology Section, Ontario Ministry of the Environment.

4. Criteria

The Ontario Ministry of the Environment (MOE) Bacterial Criteria for various water uses are presented in Table I. One of the prime reasons for inclusion of bacteriological parameters in water quality analysis is to indicate the presence of fecal contamination and thus the possible presence of pathogenic bacteria. Since the determination of specific pathogens in water is generally slow, laborious and uneconomical, specific groups of bacteria generally associated with fecal matter are used as indicators of fecal contamination.

TABLE I

Ministry of the Environment Bacterial Criteria

Maximum permissible bacterial levels in water that is to be used for:

Recreational Use

Total Coliforms 1000/100 ml Fecal Coliforms 100/100 ml Fecal Streptococci 20/100 ml

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Private Water Supplies

	To Be Treated By Chlorination Only	To Be Treated By Chlorination And Filtration
Total Coliforms Fecal Coliforms Fecal Streptococci	100/100 ml 10/100 ml 1/100 ml	400/100 ml 40/100 ml 4/100 ml
Heterotrophic Bacteria	1000/100 ml	4000/100 ml

Public Surface Water Supplies

	Receiving Full Treatment
Total Coliforms Fecal Coliforms Fecal Streptococci	5000/100 ml 500/100 ml 50/100 ml
Heterotrophic	100000/100 ml

<u>Total Coliforms:</u> This group of bacteria comprises species that are commonly associated with fecal matter (human and animal) and normal inhabitants of soil and vegetation.

Fecal Coliforms: These bacteria are mainly species associated with human and animal fecal matter and indicate a relatively recent pollution input.

Fecal Streptococci: This group of bacteria is largely associated with fecal pollution from animal and to a lesser extent man. The geometric mean of the FS results is mainly used in a ratio with the corresponding FC geometric mean (FC/FS) to gain information on the source (human or nonhuman) of pollution within areas adjacent to or at an input. If this ratio is greater than 4.0, the source of bacterial contamination is likely of human origin. If the ratio is less than 0.7 then the source is most likely nonhuman (Geldreich and Kenner, 1969). It should be noted that this ratio is used to determine the source and not the safety of the water, as animals are a potential source of organisms pathogenic to humans.

<u>Pseudomonas</u> <u>aeruginosa</u>: This bacterium is not yet an official parameter but it is a pathogen found in human fecal matter. Its presence in waters intended either for consumption or recreational use could constitute a major health hazard, as it is indicative of a local source of fecal pollution of serious concern to all users of those waters.

Heterotrophic Bacteria: This group of bacteria is indicative of general water quality and tends to reflect the level of nutrient enrichment of water.

5. Results

All survey results determined through the statistical procedures previously described are presented on graphs and maps of the survey area (Fig. 3-11, 1973; Fig. 12-19, 1974 and Fig. 20, 1969). Separate figures have been prepared for the Sanitary Indicator Bacteria levels (TC, FC and FS) and the HB and P. aer. levels. In the figures, adjoining areas or stations (Stn.) which have the same GM indicated for a parameter were homogenous for that parameter. When the term "groups" is used as a general term, it will mean both groups of stations and/or single stations that were statistically determined to delineate separate areas or locations.

All data presented in the tables and on the maps is the geometric mean. For TC, FC, FS and P. aer. the GM given is always per 100 ml while that for the HB results is per 1 ml.

I) Intensive Surveys, 1973

Sanitary Pollution Indicator Bacteria

The results from all these intensive surveys indicate that all groups were below the Ministry of the Environment Recreational Use criteria (Figs. 3, 4 and 5). There were however instances of elevated bacterial levels.

During all three surveys the water quality was within the permissible criteria for public surface water supplies and during the May survey, only lower Penetang Bay (Stn. 353, 354, 369) and the STP Effluent Discharge area (Stn. 370) in Midland Bay exceeded the desirable criteria.

With the exception of areas in Penetang (Stn. 353, 369) and Midland Bay (Stn. 370) in May and the Ontario Hospital STP (Stn. 371) in September, the water quality was within the permissible criteria for private water supplies requiring chlorination and filtration. The May survey is the only one which indicated a substantial area within the private water supply permissible criteria requiring chlorination only.

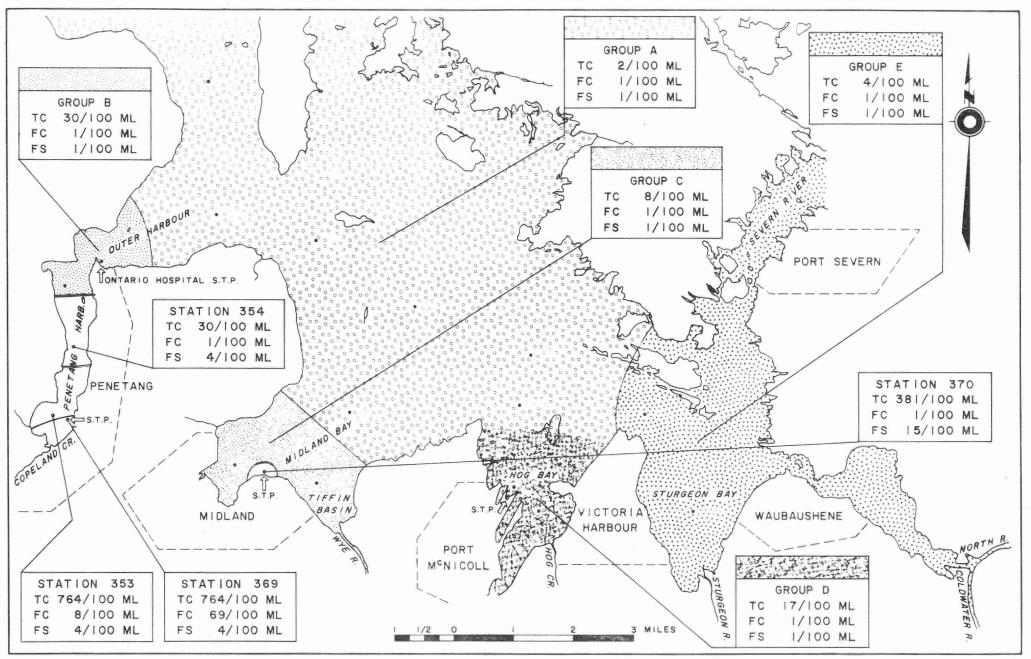


FIGURE 3 - DISTRIBUTION OF TC, FC AND FS DURING THE MAY 1973 INTENSIVE SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

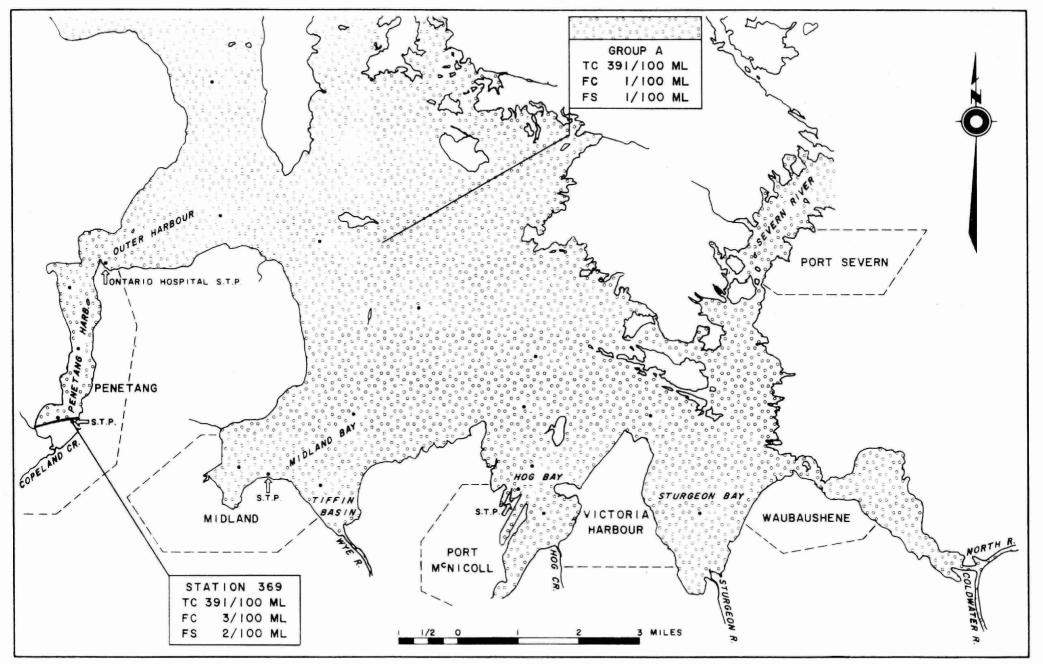


FIGURE 4 - DISTRIBUTION OF TC, FC AND FS DURING THE JULY 1973 INTENSIVE SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

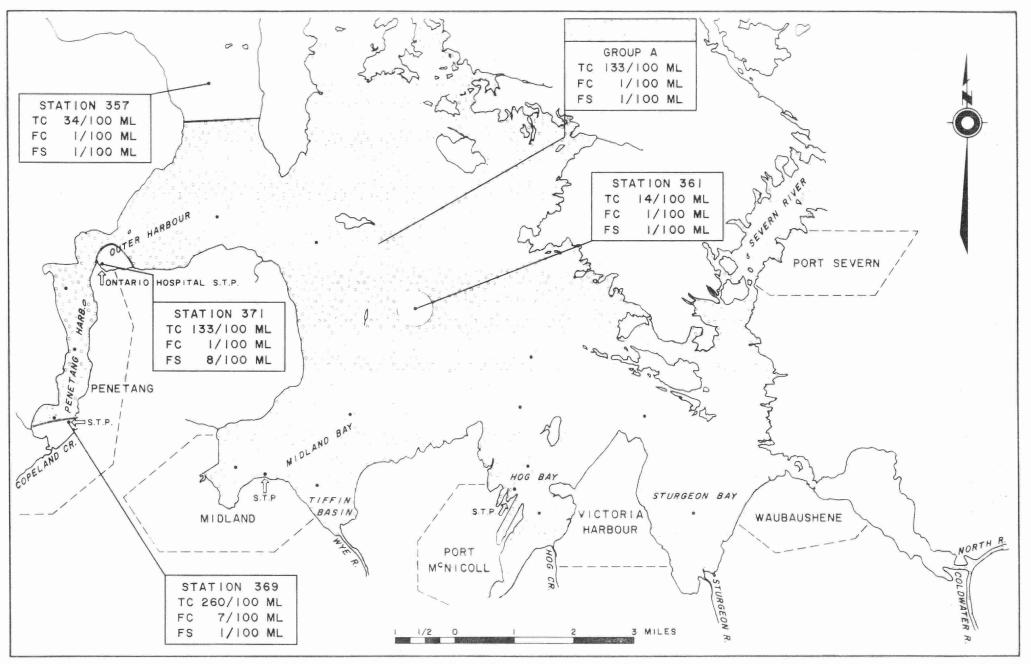


FIGURE 5 - DISTRIBUTION OF TC, FC AND FS DURING THE SEPTEMBER 1973 INTENSIVE SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

The May survey showed the area to be very heterogeneous (Fig. 3) as all four bays were nonhomogeneous with the open water area. The TC levels were elevated in the four bays, especially around the Midland and Penetang STP discharge areas, and FC levels were elevated in the area of the Penetang STP. The FC/FS ratio was intermediate over the major percentage the area, however, at the Penetang STP it was well above 4.0 and at the Midland STP it was much below 0.7.

By July (Fig. 4), almost the entire survey area formed a homogeneous group. The Penetang STP discharge area was the only one excluded due to slightly higher FC levels.

The Student-t test comparison between May and July indicated that except around the Penetang and Midland STP's, the July TC levels had increased significantly. The July FC remained at the same level except for the Penetang STP where a decrease was noted. The FS levels were significantly higher except around the Penetang STP and the Midland STP. The FC/FS ratio decreased from May and was below 0.7 for the major percentage of the area.

The September survey (Fig. 5) was somewhat less homogeneous than the July survey. The furthest nortwest area (Stn. 357) and the one near the middle of the larger basin (Stn. 361) were excluded from the larger homogeneous area because of somewhat lower TC levels and and Penetang STP (Stn. 369) which had higher TC levels. The FC/FS ratio was intermediate over most of the area except for the Penetang STP where it was above 4.0 and the Ontario Hospital STP which was below 0.7.

The Student-t test on TC results indicated a general decrease in levels from July to September, however, the levels generally were above those of the May survey. The TC levels at the Penetang STP were not significantly different from the previous surveys.

The September FS results displayed a trend similar to that of the TC results. The levels were lower than in July but still higher than in May except for the lower end of Penetang Bay and at the Ontario Hospital.

The main trends in bacterial concentrations are illustrated in Fig. 6.

II. Biweekly Surveys, 1973

Sanitary Pollution Indicator Bacteria

The results of the biweekly surveys were relatively homogeneous for each time period (Fig. 7-10). Only three areas had values above that of the major group; the Penetang STP (Stn. 369) (June to September), the Ontario Hospital STP discharge area (Stn. 371) (June), and the Midland STP (Stn. 370) (June and September).

Permissible Criteria for Public water supplies was exceeded at the Midland STP in September. In addition to the above location, the permissible criteria for private water use were exceeded in July and August by all stations. TC levels first increased and then decreased with the peak in July. This is in agreement with the intensive survey data, but the levels indicated are higher and in July the TC density exceeds the Recreational Use Criteria. FC concentrations showed a slight increase from month to month but the only significant increase was from June to September. FS levels followed a similar pattern to TC levels but the differences were too small to be significant.

The main trends are indicated in Fig. 11.

III. Intensive Surveys, 1974

A Sanitary Pollution Indicator Bacteria

This year the results indicated that almost the entire survey area exceeded Recreational Use Criteria and Permissible Private Water Supply Criteria during the July survey. The Public Surface Water Supply criteria was not exceeded during any of the surveys.

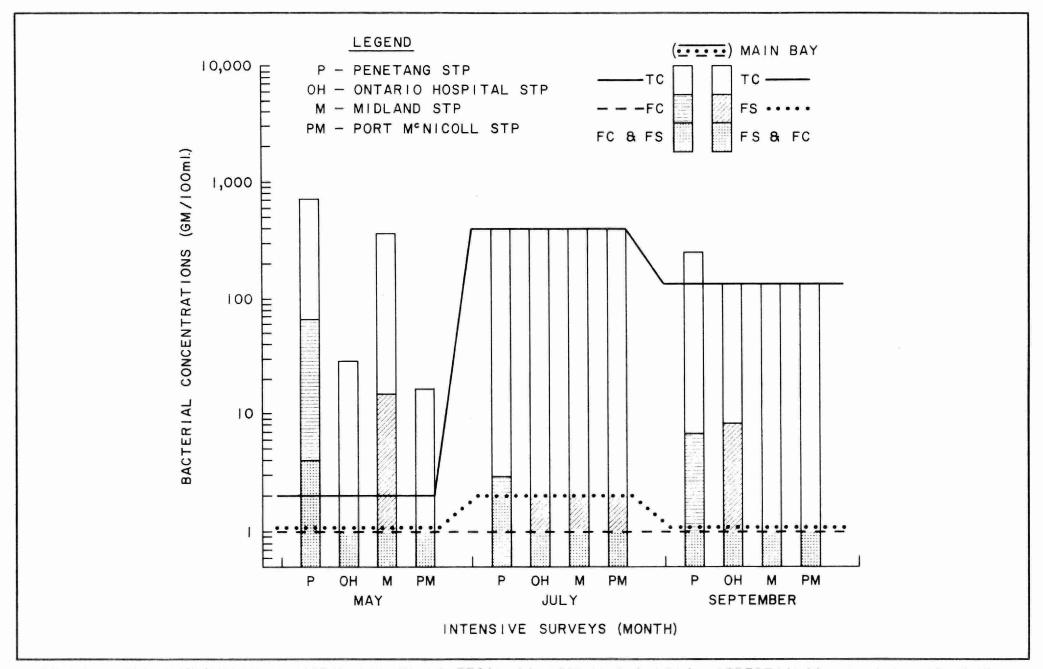


FIGURE 6 - CONCENTRATIONS OF TOTAL COLIFORMS, FECAL COLIFORMS AND FECAL STREPTOCOCCI DURING THE 1973 INTENSIVE SURVEYS FOR THE MAIN BAY AND STP EFFLUENT DISCHARGE AREAS.

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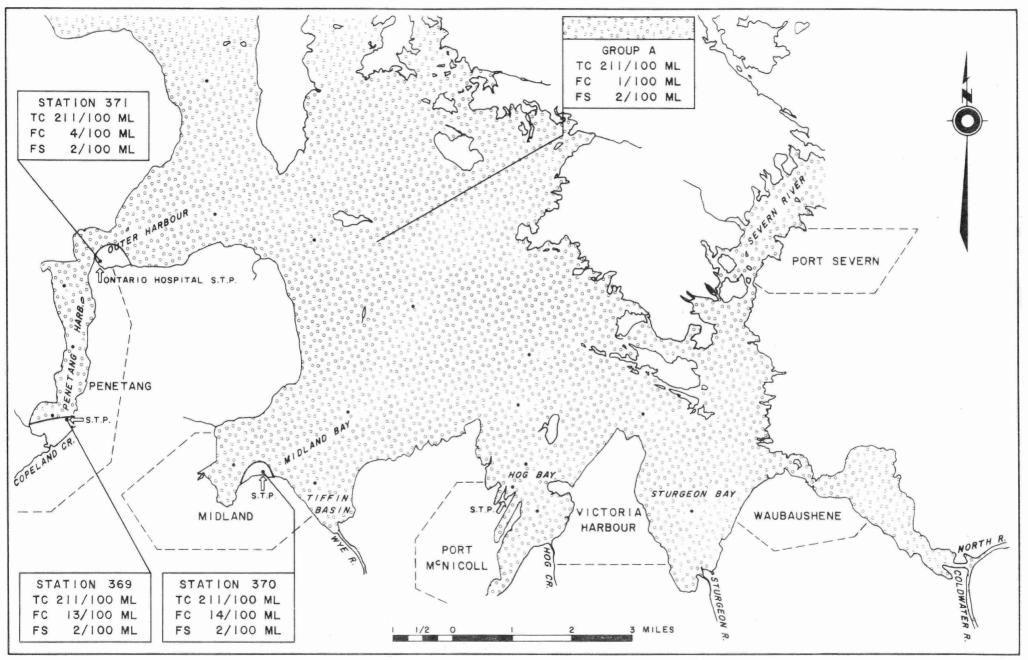


FIGURE 7 - DISTRIBUTION OF TC, FC AND FS DURING THE JUNE 1973 BI-WEEKLY SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

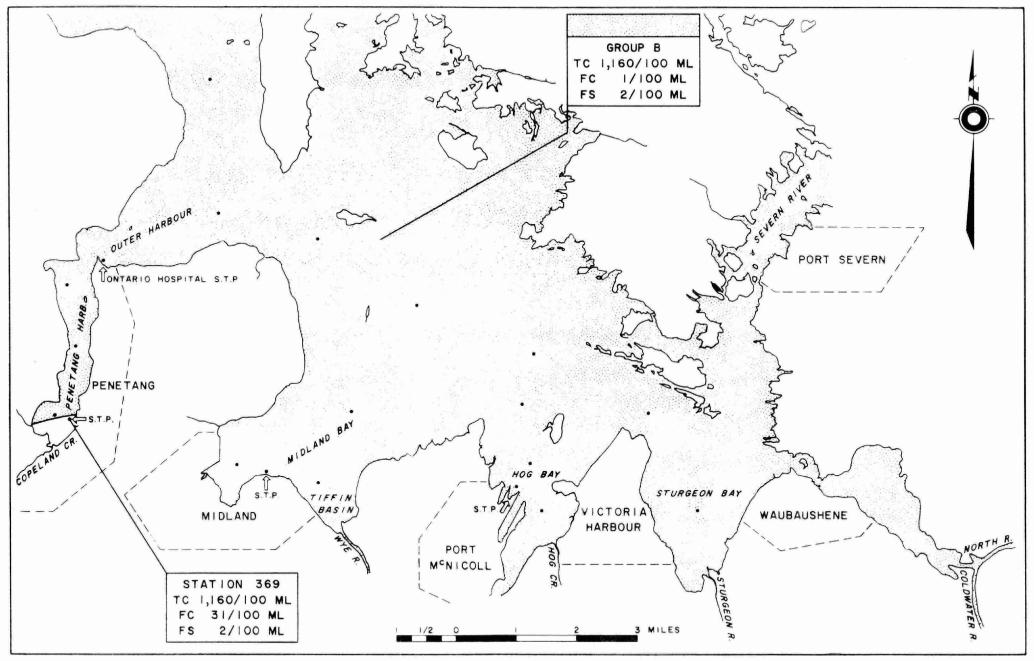


FIGURE 8 - DISTRIBUTION OF TC, FC AND FS DURING THE JULY 1973 BI-WEEKLY SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

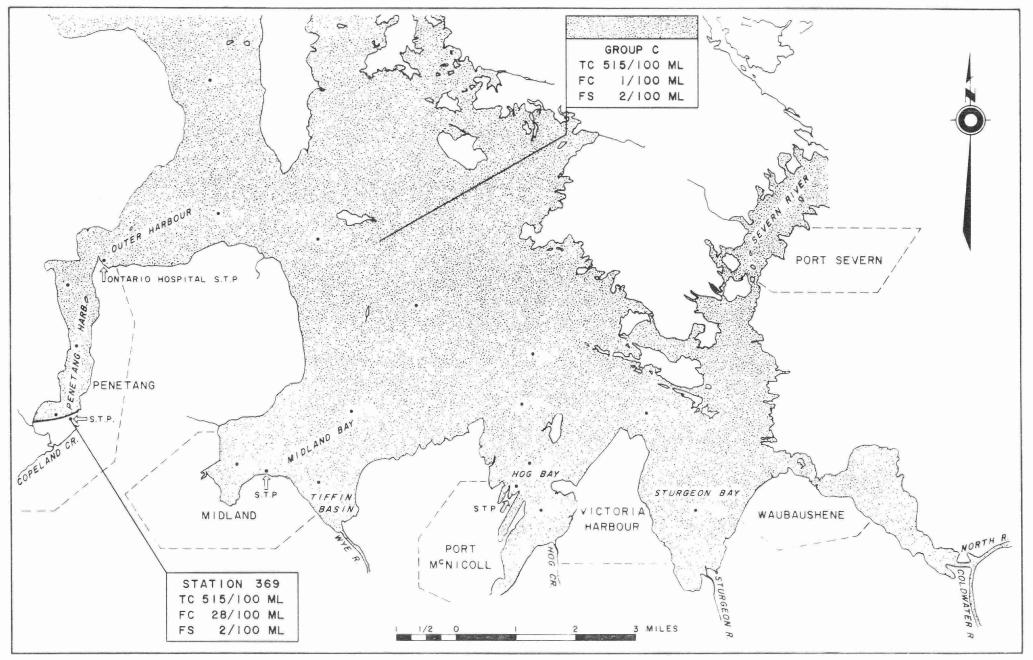


FIGURE 9 - DISTRIBUTION OF TC, FC AND FS DURING THE AUGUST 1973 BI-WEEKLY SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

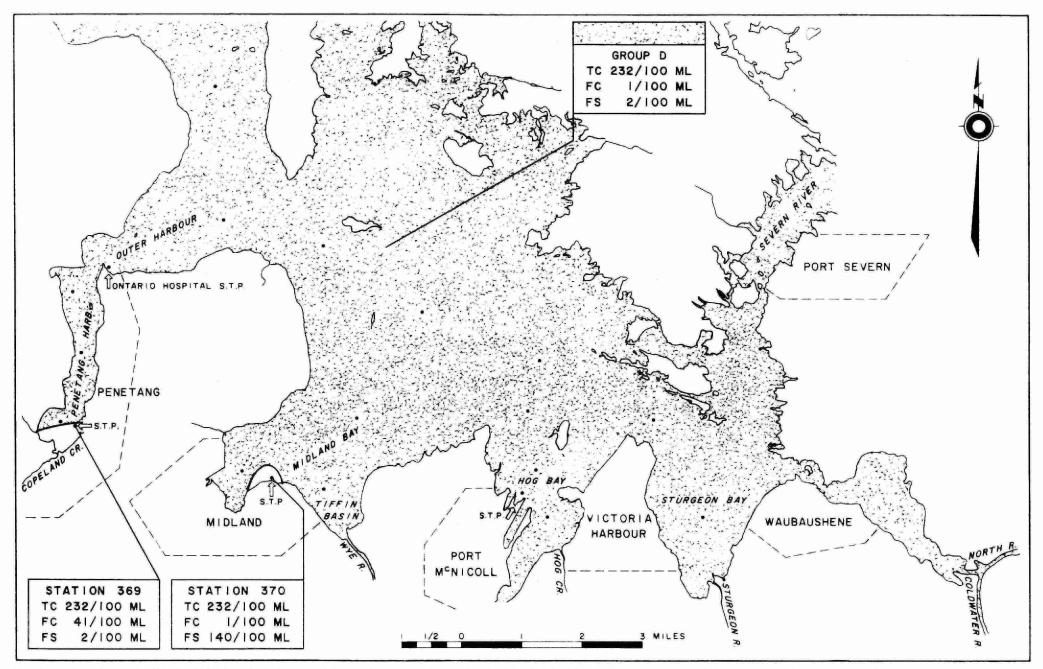


FIGURE 10 - DISTRIBUTION OF TC, FC AND FS DURING THE SEPTEMBER 1973 BI-WEEKLY SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

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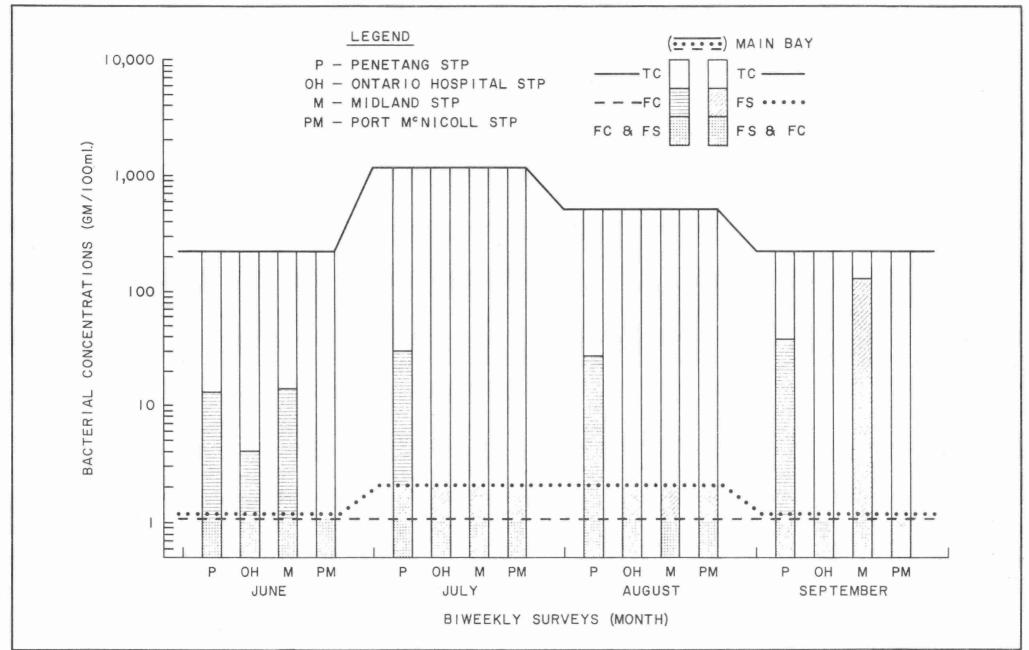


FIGURE 11 - CONCENTRATIONS OF TOTAL COLIFORMS, FECAL COLIFORMS, AND FECAL STREPTOCOCCI DURING THE 1973 BIWEEKLY SURVEYS FOR THE MAIN BAY AND STP EFFLUENT DISCHARGE AREAS.

The June results (Fig. 12) were homogeneous for the entire area and water quality was very good. The only exception was in western Midland Bay (Stn. 358) where a slightly elevated TC level was observed. The FC/FS ratio was 0.7 for the entire area surveyed.

In July (Fig. 13), Penetang Bay had significantly higher levels of FC than the remainder of the survey area and an FC/FS ratio exceeding 4.0. The entire area had significantly higher TC levels than in June, while FC and FS levels had decreased slightly. One exception was in Penetang Bay where FC densities had increased. Sturgeon Bay (Stn. 365) was the only area with TC concentrations below acceptable criteria.

Analysis of the September data (Fig. 14) showed a more heterogeneous situation than in June or July. The lower end of Penetang and Hog Bays, Sturgeon Bay and the western side of Midland Bay all had poorer water quality than the remainder of the survey area. In particular, the Penetang and Midland STP discharge areas had elevated levels of all three parameters. All TC concentrations were decreased from July though they had not yet returned to those of June. The FC densities were generally the same as July with only slight elevations in western Midland Bay to a level higher than that observed in June and a decrease in one area of Penetang Bay (Stn. 354). FS concentrations were somewhat increased with the FC/FS ratio above 4.0 only in one location in Southern Penetang Bay (Stn. 353). The ratio at the Penetang and Midland STP's was below 0.7.

Comparisons of bacteriological densities for the three surveys are presented in Fig. 15.

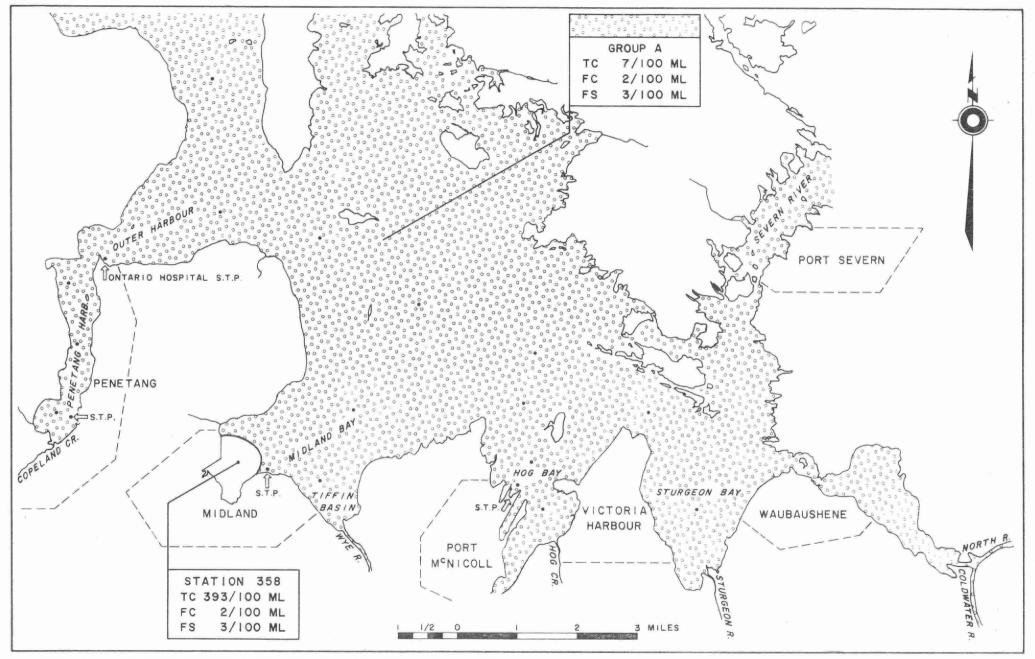


FIGURE 12 - DISTRIBUTION OF TC, FC AND FS DURING THE JUNE 1974 INTENSIVE SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

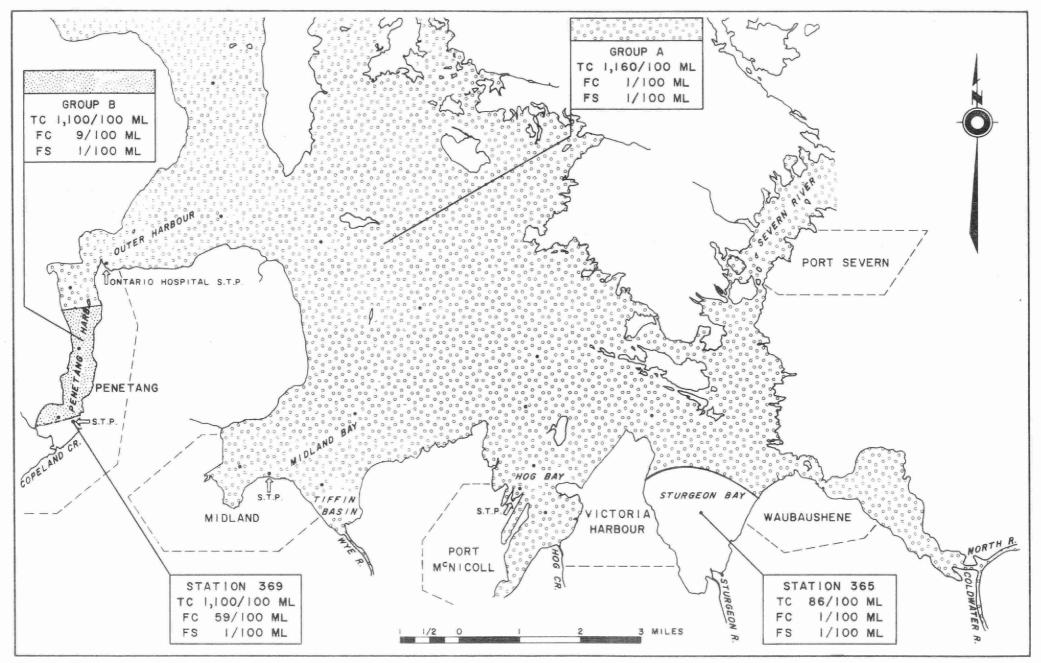


FIGURE 13 - DISTRIBUTION OF TC, FC AND FS DURING THE JULY 1974 INTENSIVE SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

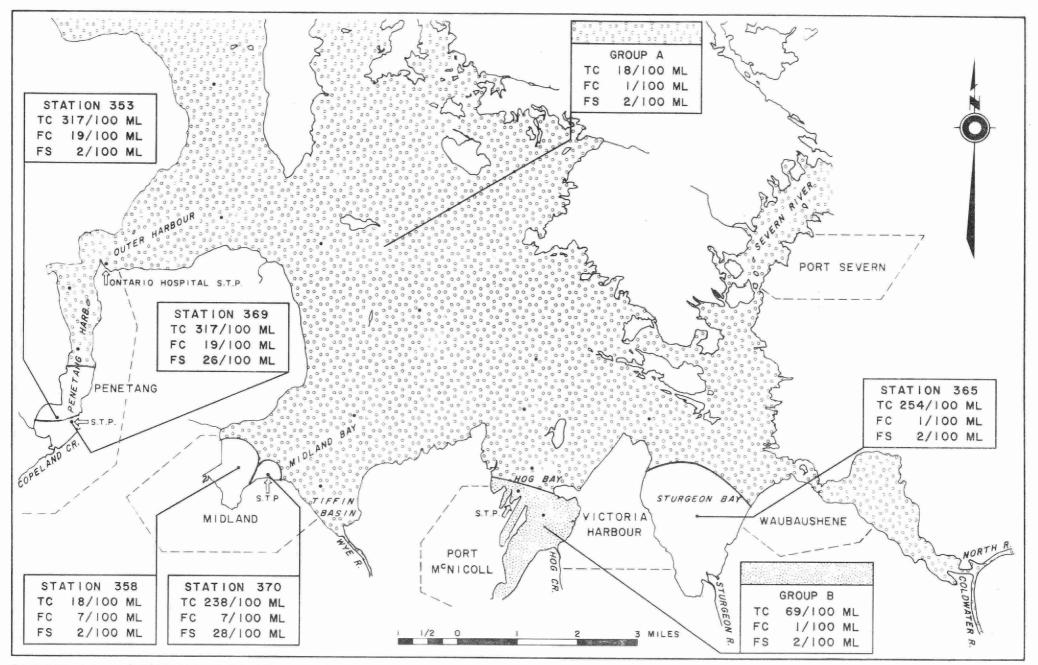


FIGURE 14 - DISTRIBUTION OF TC, FC AND FS DURING THE SEPTEMBER 1974 INTENSIVE SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

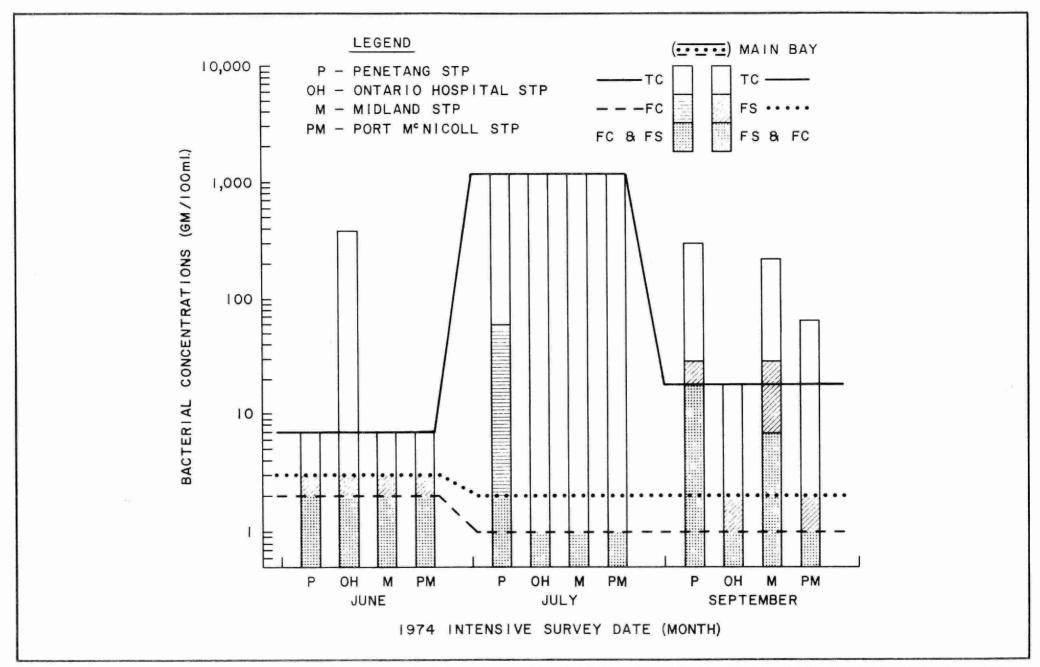


FIGURE 15 - CONCENTRATIONS OF TOTAL COLIFORMS, FECAL COLIFORMS AND FECAL STREPTOCOCCI DURING THE 1974
INTENSIVE SURVEYS FOR THE MAIN BAY AND STP EFFLUENT DISCHARGE AREAS.

B Heterotrophic Bacteria and Pseudomonas aeruginosa

In June (Fig. 16) the entire area with the exception of elevated HB levels in western Midland Bay (Stn. 358) was homogeneous for both HB and P.aer.

The July survey (Fig. 17) indicated an overall significant increase in HB levels from June. This was particularly true in Penetang and Midland Bays and around the Port McNicoll STP. The only area showing a significant increase in P.aer. densities was the Penetang STP discharge area.

Analysis of the September results (Fig. 18) indicated that only lower Midland Bay had elevated HB levels, the remainder of the survey area was homogeneous. Lower Midland Bay was the only area that did not have at least a slight decrease in HB levels between July and September, however, the decreases in level were only significant in Penetang Bay and around the Port McNicoll STP. P.aer. concentrations remainded unchanged except in the Penetang STP discharge area where they decreased to a background level.

Overall trends are illustrated in Fig. 19.

4. Comparison of 1973, 1974 and 1969 Intensive Surveys

A 1973 and 1974

The June 1974 survey data has only a maximum of three pieces of information, thus the validity of a comparison to the 1973 intensive survey is uncertain. However, it appears that the 1974 survey took place after spring mixing had occurred whereas the 1973 survey had taken place before turnover. This may have led to the more homogeneous data in 1974. The TC levels were up slightly in the open water area from 1973 but down in Penetang Bay. The remainder of the area was not significantly different from the previous year. The FC and FS concentrations were higher through most of the area in 1974 except at the south end of Penetang Bay which was lower.

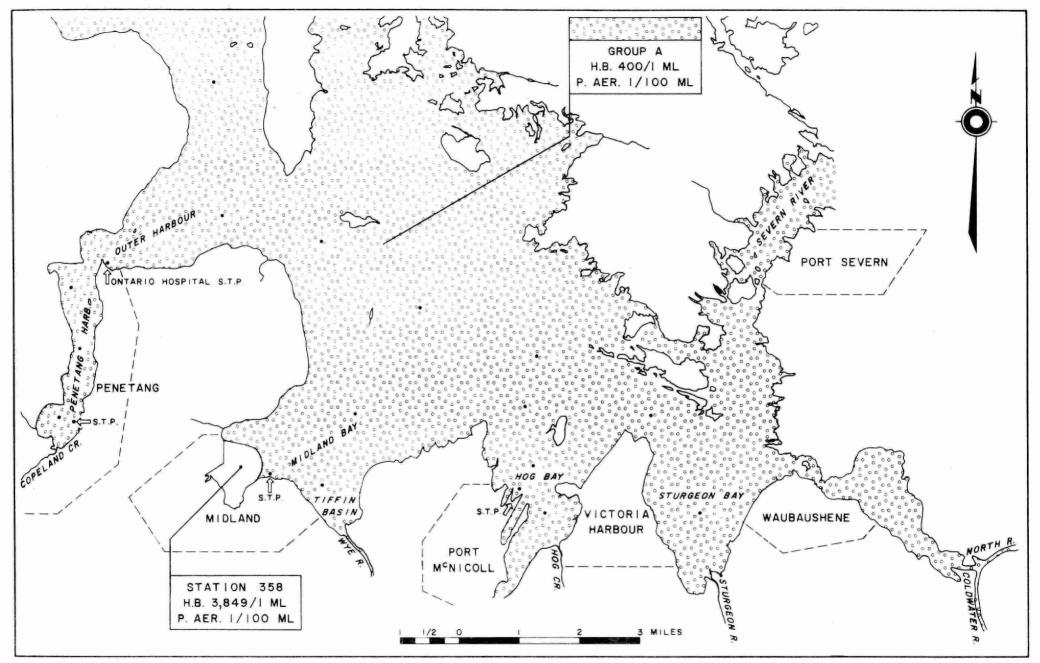


FIGURE 16 - DISTRIBUTION OF H.B. AND P. AER. DURING THE JUNE 1974 INTENSIVE SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

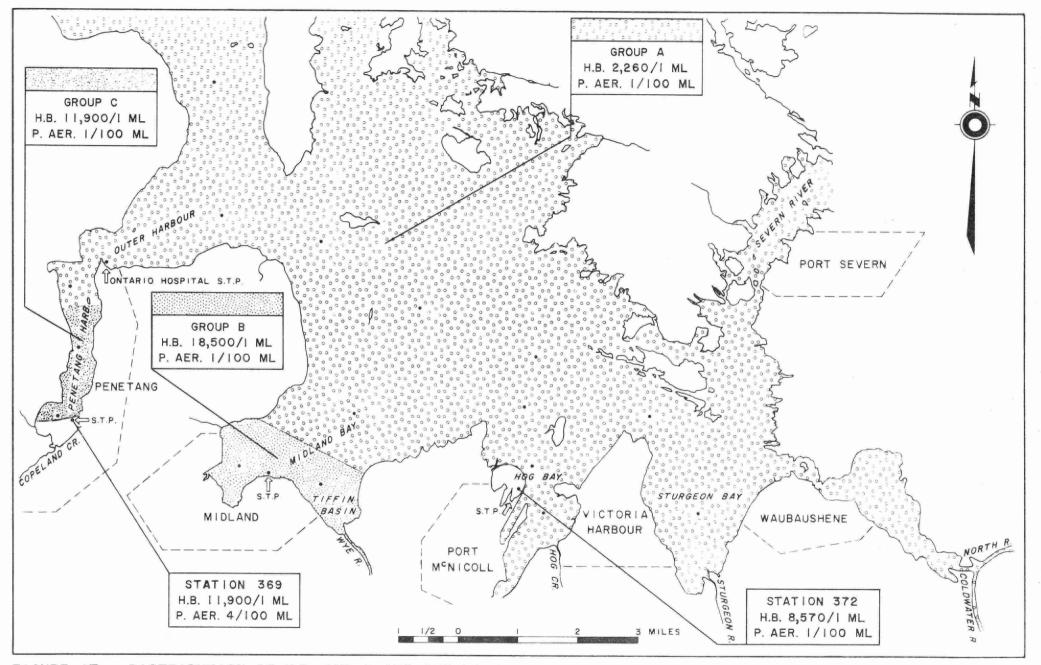


FIGURE 17 - DISTRIBUTION OF H.B. AND P. AER. DURING THE JULY 1974 INTENSIVE SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

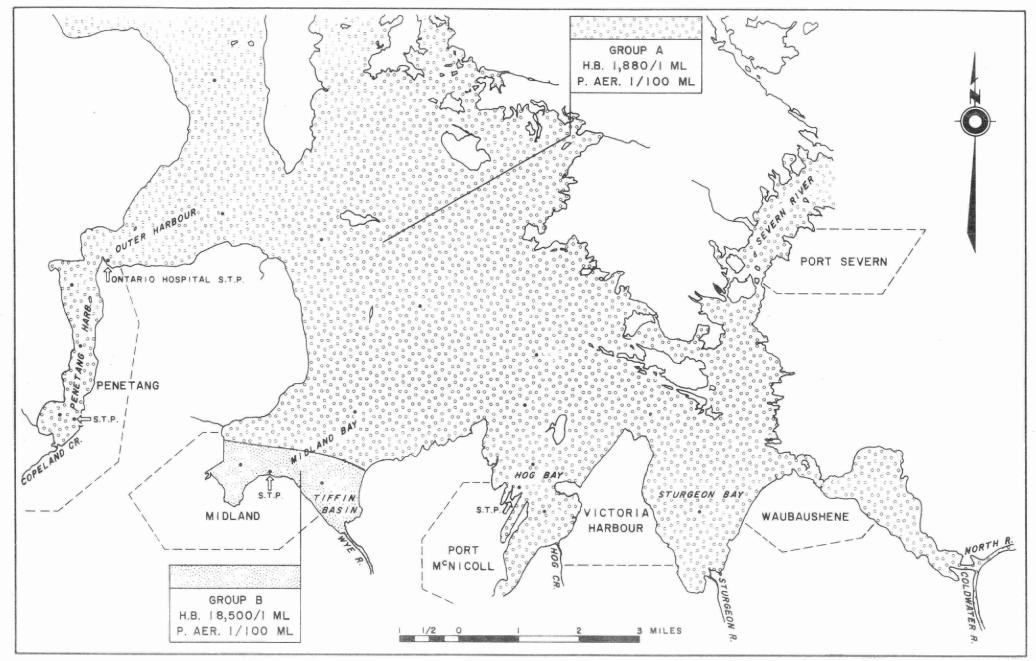


FIGURE 18 - DISTRIBUTION OF H.B. AND P. AER. DURING THE SEPTEMBER 1974 INTENSIVE SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

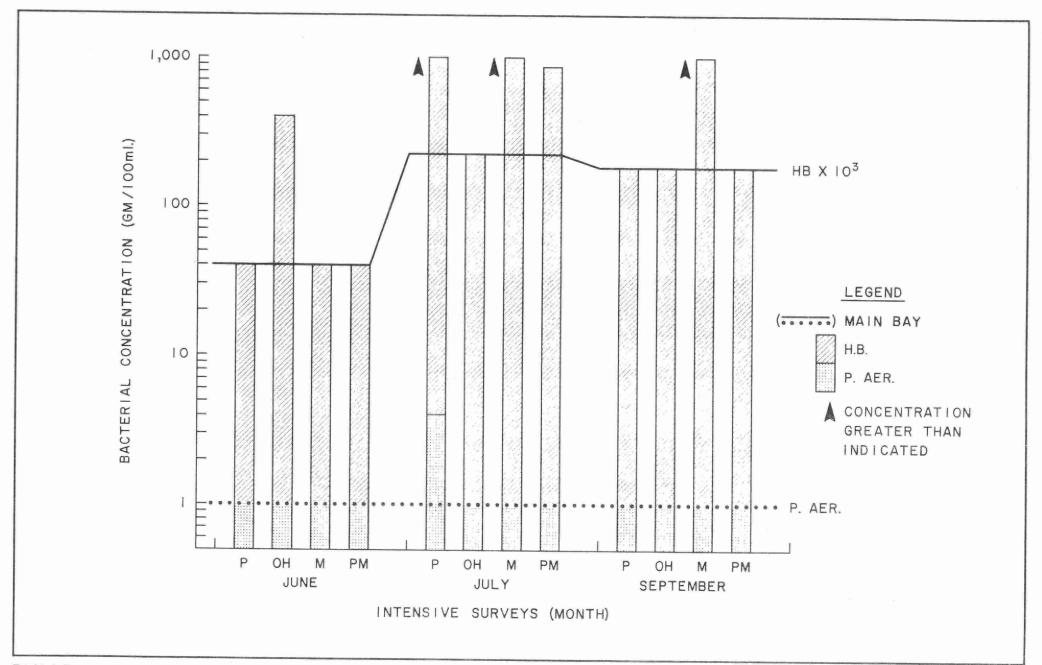


FIGURE 19 - CONCENTRATIONS OF HETEROTROPHIC BACTERIA AND PSEUDOMONAS AERUGINOSA DURING 1974 INTENSIVE SURVEYS FOR THE MAIN BAY AND S.T.P. EFFLUENT DISCHARGE AREAS.

Total coliform densities in July 1974 were significantly higher than in July 1973. Fecal coliform densities were at the same level except in Penetang Bay which were higher in 1973. The FS concentrations were slightly lower in 1974.

The September results showed no significant difference in TC densities in the four Bays, however, the remainder of the survey area had lower TC levels in 1974. There was little difference in the FC levels except for somewhat higher levels around the Midland STP in 1974. Fecal streptococci levels were increased in 1974 throughout the survey area.

B 1969, 1973 and 1974 July Surveys

During the July 1969 survey (Fig. 20) Penetang Bay and the Midland STP discharge area had elevated FC and FS levels. The Penetang STP area also had elevated TC densities. The TC densities over the entire area exceeded recreational use and private water supply criteria and were significantly higher than those of both 1973 and 1974. FC levels in 1969 were higher than 1973 in Penetang Bay but otherwise densities were the same as those of 1973 and 1974 for the majority of the area. FS levels for 1969 were generally intermediate between 1973 and 1974.

The inputs from the two STP's existing in 1969 (Penetang and Midland) were compared with levels observed in 1973 and 1974. The Penetang STP discharge area (Stn. 369) had 1969 TC and FC levels that were significantly higher than those of 1973 and 1974. The FS levels remained at approximately the same concentration. The Midland STP discharge area had 1969 TC densities greater than those observed in 1973. All other parameters showed no significant changes.

Fig. 21 compares the bacterial levels observed during the July surveys of 1969, 1973 and 1974.

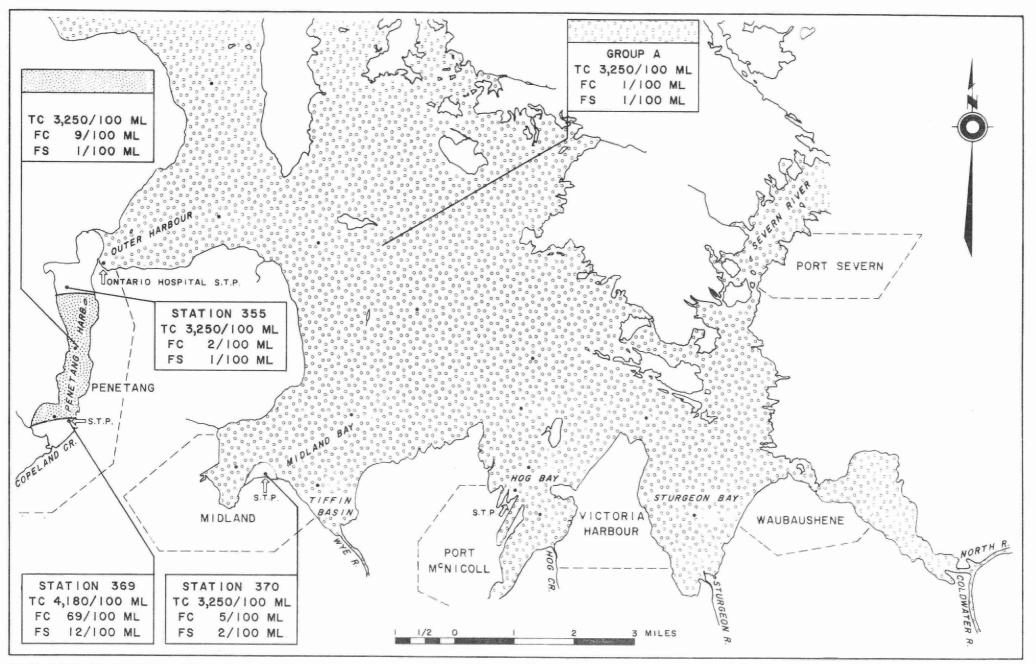


FIGURE 20 - DISTRIBUTION OF TC, FC AND FS DURING THE JULY 1969 INTENSIVE SURVEY OF THE PENETANGUISHENE-WAUBAUSHENE AREA.

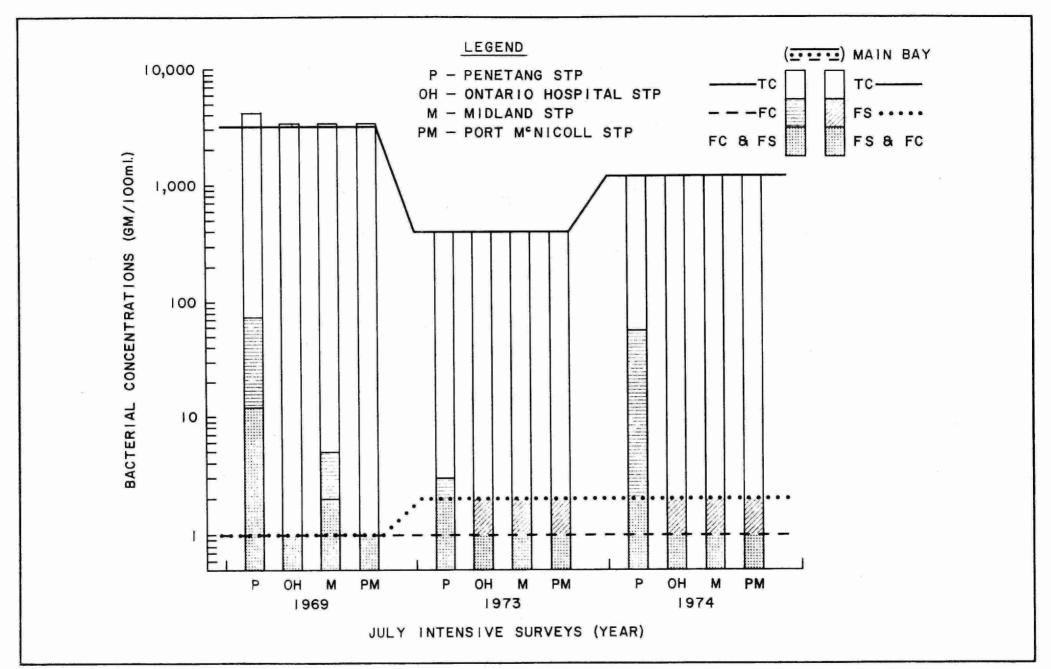


FIGURE 21 - COMPARISON OF LEVELS OF TOTAL COLIFORMS, FECAL COLIFORMS AND FECAL STREPTOCOCCI DURING THE 1969, 1973 AND 1974 JULY INTENSIVE SURVEYS FOR THE MAIN BAY AND STP EFFLUENT DISCHARGE AREAS.

AREAS.

Discussion

The bacterial water quality of the Penetang-Waubaushene Survey area is reasonably variable. Levels of sanitary indicator bacteria are acceptable for recreational and public surface water supplies during the spring and fall periods. However, increased summer populations and activity appears to have a detrimental effect on the area and the "recreational use" criteria can be exceeded. This was evidenced by the results from the 1973 July biweekly and 1974 July intensive surveys. There is no direct measurement of the input created by the increased summer recreational activity, however, peak bacterial levels were apparent over the entire area during the summer. The contribution of effluent inputs was apparent from the number of times that significantly elevated bacterial levels were present in the vicinity of the STP's (Fig. 22). The major input of this form was at the Penetang STP which had significantly higher bacterial densities during nine of the ten survey periods. The next largest contribution was from the Midland STP.

Problems with waste treatment at the Penetang STP are further illustrated by the FC/FS ratios (Geldreich and Kenner, 1969) within the four bays (Fig. 23). The only area with ratios indicating possible human fecal contamination was the lower end of Penetang Bay. This occurred six times during the ten survey periods. There was a continual decrease in the ratio with increasing distance from this STP. An interesting finding was that the FC/FS ratio of the Midland STP effluent was indicative of a nonhuman input. This is possibly explained by the fact that the Midland STP had a hydraulic loading problem caused by a combined sewer system and groundwater seepage (Marton, 1975). In addition, problems were experienced with input from industrial wastes, such as a metal polishing plant, which could alter the ratio. A possible indication of the improvement in the waste treatment at Port McNicoll is the decrease in FC/FS ratios from 1973 to 1974. In

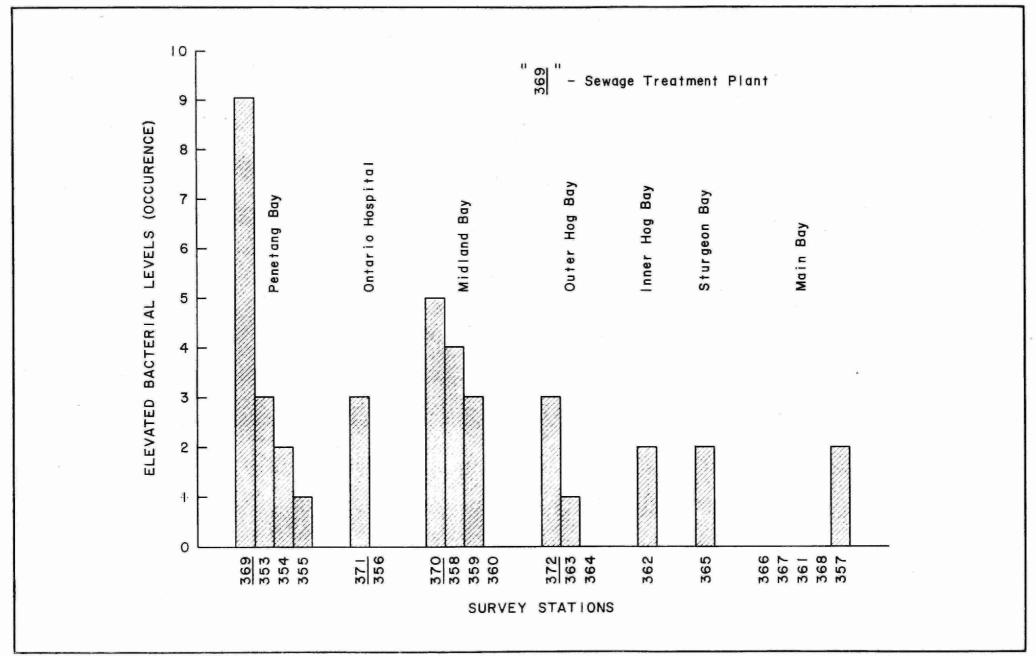


FIGURE 22 - OCCURENCE OF ELEVATED BACTERIAL LEVELS DURING THE PENETANGUISHENE-WAUBAUSHENE SURVEYS.

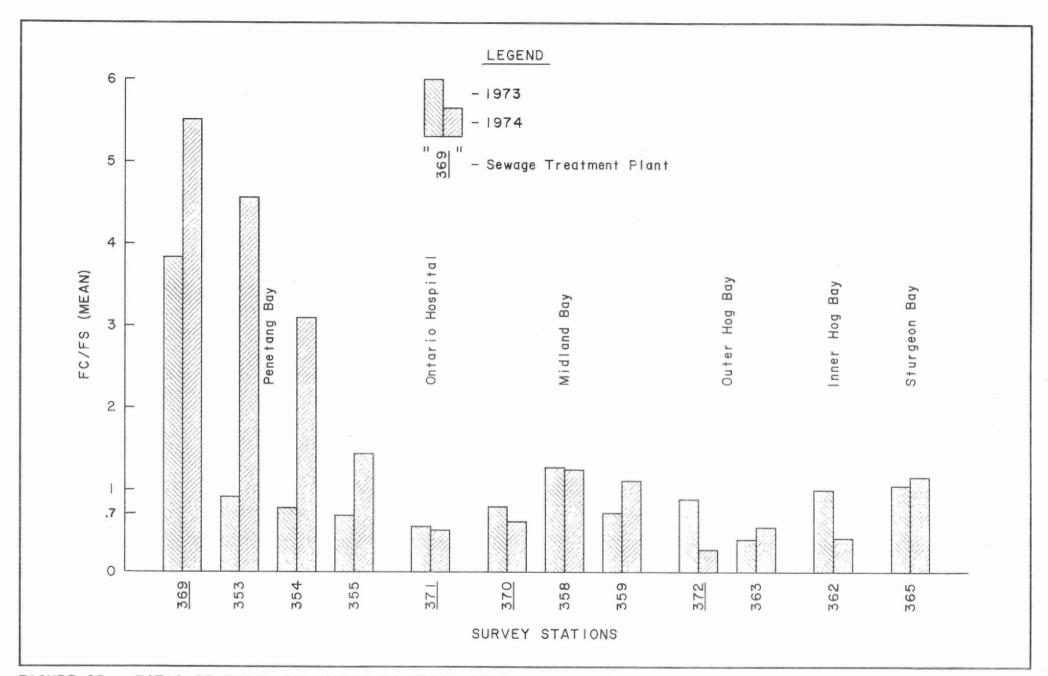


FIGURE 23 - RATIO OF FECAL COLIFORMS TO FECAL STREPTOCOCCI FROM 1973 AND 1974 INTENSIVE SURVEYS.

1973, Port McNicoll was on individual septic tanks and the ratios were generally at intermediate levels. In 1974, a number of homes were hooked up to the STP and ratios were indicative of nonhuman inputs.

The <u>Pseudomonas aeruginosa</u> levels provide still another indicator of waste treatment problems at the Penetang STP (Fig. 24). In July 1974, the Penetang STP effluent area was the only area with P. aer. densities significantly higher than the remainder of the survey area.

Although water quality had improved from 1969 to 1973 and 1974, the reasons for the improvement were not readily apparent as waste treatment in the area had not changed substantially. Contributing factors may be:

- a) the level of Lake Huron was lower by about one foot in 1969 (Department of Transport 1969, 1973 and 1974).
- b) greater precipitation preceding and during the 1969 survey. In fact there was no precipitation during the 1973 and 1974 surveys (Department of the Environment 1970, 1973 and 1974).
- c) higher bacterial levels in the Penetang and Midland STP effluents in 1969 possibly due to bypassing on rainy days.

Hopefully, the improvements in waste treatment in the area will result in better quality effluents and an improvement in the general water quality of the area. Improvement is definitely required as this is a major summer recreation area. There are many cottages located along its shores, a number of historic sites that draw tourists, and there is ready access to the water for swimming and boating. Many of the summer inhabitants use the water not only for body contact sports but also for drinking, unfortunately often without treatment. Although the FC and FS concentrations are not exceedingly high, except in the vicinity of the Penetang and



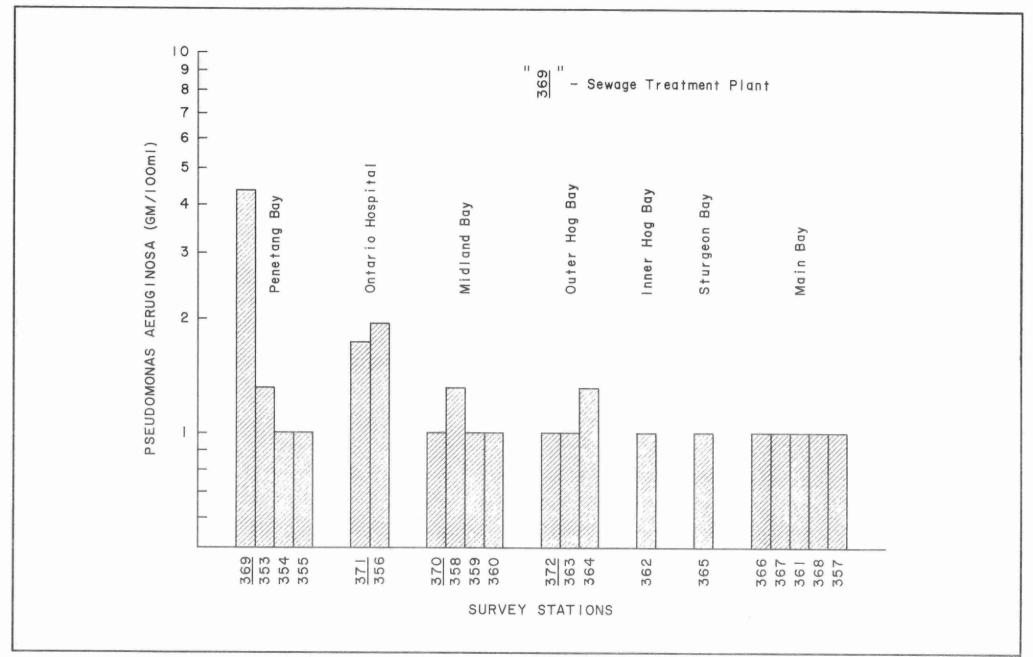


FIGURE 24 - GEOMETRIC MEAN LEVELS OF PSEUDOMONAS AERUGINOSA DURING THE JULY 1974 INTENSIVE SURVEY.

Midland STP, the generally high bacterial densities as indicated by TC and HB levels are cause for concern because of the nature of the area. It should be further noted that most of the sampling locations (except for the STP's) were located well offshore, thus the bacterial quality of the nearshore water actually used for swimming and drinking was unknown and could have been worse. One area in particular which should be examined more closely is the trailer park just south of the Penetang STP.

The stress in studies like these should be on "intensive" surveys rather than single sampling or a biweekly type of survey. A plot of the TC levels for the Penetanguishene-Waubaushene survey area over the five intensive surveys indicates the type of variability in levels that can occur (Fig. 25).

The intensive surveys permitted the use of a statistical analysis which provided the means for zoning areas with elevated bacterial levels and determining the geometric means of these levels and the degree of variation around these means. A single sample would have provided only a count which in most cases would not have been representative of the water quality. manipulation, the biweekly sampling provided enough data for a statistical analysis though not as much as the intensive surveys, and it was not as sensitive to geographical heterogeneity in bacterial levels as the intensive surveys. In addition, the bacterial levels indicated by the biweekly surveys frequently differed from those of the intensive surveys, particularly the TC levels which were considerably higher during the biweekly surveys. This was possibly due to the fact that the biweekly results were a composite of the results of a series of single samples with all their related problems. If the variation in TC levels during the 1973 intensive surveys was representative of the weekly variation during the biweekly surveys, then the dates of the biweekly sampling generally correspond with days when TC levels were high.

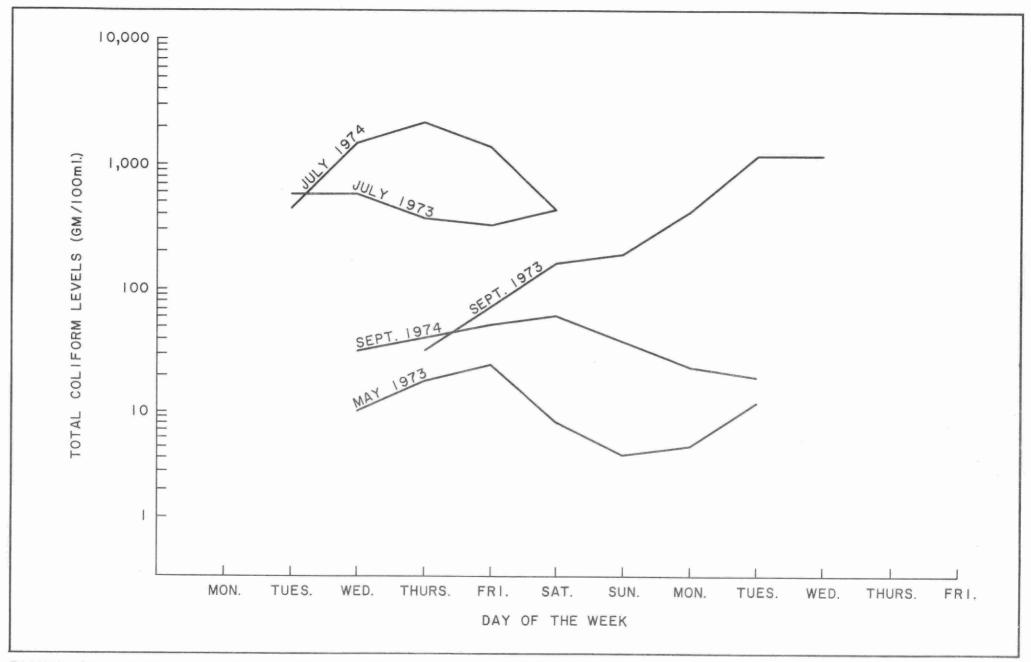


FIGURE 25 - OVERALL TOTAL COLIFORM LEVELS (GM/IOOMI) DURING 1973 AND 1974 INTENSIVE SURVEYS.

Summary and Recommendations

In conclusion, the 1973-74 surveys indicated that the bacterial quality of the water in the Penetanguishene-Waubaushene area is not satisfactory particularly in view of its use during the summer. It would be wise to make a thorough analysis of drinking water drawn from the bays, particularly by cottages and of known swimming areas, such as the trailer park, located just south of the Penetang STP.

The water quality problems in the area are caused at least in part by the STP's, particularly the Penetang STP. Water Quality may be better in the future as a result of the improvements being made to the waste treatment facilities in the future. After time has been allowed for the improvements to have effect in the area, another set of intensive surveys should be carried out.

Finally, a comparison of the intensive and biweekly type of survey methods strengthened our view that the intensive method of surveying was superior.

REFERENCES

- Bousfeld, J.I., Smith, L.G. and Trueman, W.R. 1973. The use of semiautomated pipettes in the viable counting of bacteria. J. Appl. Bact. 36:297-299.
- Department of the Environment, Canada, July 1970, 1973 and 1974. Monthly Water Level Bulletin, Great Lakes and Montreal Harbour. Inland Waters Directorate.
- Department of Transport, Canada, June and July, 1969, 1973 and 1974.
 Monthly Record, Meteorological Observations in Canada. Meteorological Branch.
- 4. Foot, H.C. and Taylor, B.C. 1949. The influence of the composition of the medium on the growth of bacteria from water. Proceedings of the Society for Applied Bacteriology, pp. 11-13.
- 5. Geldreich, E.E. and Kenner, B.A. 1969. Concepts of fecal streptococci in stream pollution. Journal W.P.C.F. 41, R336-R352.
- Jones, Moya. 1970. The Georgian Bay survey bacteriological report.
 Ontario Water Resources Commission, Laboratory Division. 45 pp.
- 7. Levin, M. A. and Cabelli, V.J. 1972. Membrane filter techniques for enumeration of Pseudomonas aeruginosa. Appl. Microbiol. 24, 864-870.
- Marton, F. 1975. Personal Communication. Ministry of the Environment,
 Central Region.
- Veal, D.M. and Michalski, M.F.P. 1971. A case of nutrient enrichment in an inshore area of Georgian Bay. Proceedings 14th Conference Great Lakes Research, 277-292.

REFERENCES

- Bousfeld, J.I., Smith, L.G. and Trueman, W.R. 1973. The use of semiautomated pipettes in the viable counting of bacteria. J. Appl. Bact. 36:297-299.
- Department of the Environment, Canada, July 1970, 1973 and 1974. Monthly Water Level Bulletin, Great Lakes and Montreal Harbour. Inland Waters Directorate.
- Department of Transport, Canada, June and July, 1969, 1973 and 1974.
 Monthly Record, Meteorological Observations in Canada. Meteorological Branch.
- 4. Foot, H.C. and Taylor, B.C. 1949. The influence of the composition of the medium on the growth of bacteria from water. Proceedings of the Society for Applied Bacteriology, pp. 11-13.
- 5. Geldreich, E.E. and Kenner, B.A. 1969. Concepts of fecal streptococci in stream pollution. Journal W.P.C.F. 41, R336-R352.
- Jones, Moya. 1970. The Georgian Bay survey bacteriological report.
 Ontario Water Resources Commission, Laboratory Division. 45 pp.
- 7. Levin, M. A. and Cabelli, V.J. 1972. Membrane filter techniques for enumeration of Pseudomonas aeruginosa. Appl. Microbiol. 24, 864-870.
- Marton, F. 1975. Personal Communication. Ministry of the Environment,
 Central Region.
- Veal, D.M. and Michalski, M.F.P. 1971. A case of nutrient enrichment in an inshore area of Georgian Bay. Proceedings 14th Conference Great Lakes Research, 277-292.

A BACTERIOLOGICAL STUDY OF THE PENETANGUISHENE-WAUBAUSHENE AREA OF GEORGIAN BAY - 1976

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ABSTRACT

Surveys of Georgian Bay in the vicinity of Penetanguishene-Waubaushene were conducted in 1976 as part of a continuous program to monitor water quality and the effects of improvements carried out on waste treatment facilities in the area. The results showed that the levels of sanitary indicator bacteria were, with a few exceptions, within the acceptable limits for recreational use and private water supplies requiring chlorination and filtration. This represents an improvement in the water quality since 1973.

A BACTERIOLOGICAL STUDY OF THE PENETANGUISHENE-WAUBAUSHENE AREA OF GEORGIAN BAY 1976

INTRODUCTION

A survey of the Penetanguishene-Waubaushene area of Georgian Bay was conducted in 1976 as part of a continuing Ministry of the Environment (MOE) program to monitor Great Lakes water quality. The survey was designed to determine the water quality of the area with respect to MOE Bacterial Criteria for various water uses and International Joint Commission objectives, to observe the influence of sewage treatment plant (STP) effluents and to monitor the effect of improvements carried out on waste treatment facilities in the area.

A description of the survey area and the location of the sampling stations may be found in the Penetang-Waubaushene 1973-1974 report (2).

At the time of the 1976 survey, a number of improvements had been made to the waste treatment facilities in the area. Phosphate removal at the Ontario Hospital and Penetang STP's was fully operational with phosphate levels of 1 ppm or less in the effluents. The Midland STP was achieving 80% phosphate removal although levels in the effluent were still greater than 1 ppm. Port McNicoll had completed conversion from septic tank tile systems to a sewage collector system combined with a secondary treatment plant. Victoria Harbour and Waubaushene remained on individual septic tank systems.

METHODS

1. Field Procedures

Bacteriological samples were collected at the 20 stations used in the 1973-74 surveys. Biweekly sampling was carried out from April 29, 1976 to October 5, 1976 with the exception of the week of September 19 to 25.

Samples were collected in sterile, 175 ml glass bottles at a depth of approximately 1.5 m below the surface. The samples were immediately iced and shipped to the MOE Toronto Laboratory for analysis.

2. Laboratory Procedures

All samples were analyzed for Total Coliform (TC), Fecal Coliform (FC) and Fecal Streptococci (FS). Membrane filtration analyses were conducted according to Standard Methods (13th edition) using m-Endo Agar LES (Difco) for TC, MacConkey Membrane Broth (Oxoid) for FC and M-Enterococcus Agar (Difco) for FS (2).

All samples were also analyzed for Heterotrophic Bacteria (HB) and Pseudomonas aeruginosa (P. aer.). The HB method consisted of a spot plate technique on CPS Agar (3) at 20°C for seven days. The P. aer. method utilized was the membrane filtration procedure of Levin and Cabelli (mPA). The incubation was at 41.5°C for 48 hours (4). Analysis of all samples were conducted within 24 hours of sampling.

3. Statistical Methods

The total coliform and heterotrophic bacteria results were organized into five overlapping time groups, April 27 to June 8 (May survey), May 26 to July 8 (June survey), June 23 to August 4 (July survey), July 21 to September 1 (August survey) and August 18 to October 5 (September survey). The log geometric mean, the variance and the standard error were calculated for each time period to obtain the group statistics at each station. The analysis of variance (ANOVA) and Bartlett's chi-squared test of homogeneity were performed as during the Penetang-Waubaushene 1973-74 survey (1).

The total coliform results were subjected to this statistical analysis twice. The first time, results below the detectable limit were omitted. The second time, these results were included as a maximum count (e.g. a result of less than 4 TC per 100 ml was included as 4 TC per 100 ml). Additional Students-t-tests were used to compare the 1976 surveys to the 1973 biweekly sampling. The amount of data available for FC, FS and P. aer. was insufficient for the above statistical analysis because concentrations were below detection levels (less than 4/100 ml). Analysis of P. aer. was carried out at the STP stations 6 times and only twice at other stations. A list of stations where FC, FS and P. aer. were detected was compiled for the study period (Tables I and II).

CRITERIA

The Ministry of the Environment Bacterial Criteria and/or comments about TC, FC, FS, P. aer. and heterotrophic bacteria may be found in the Penetang-Waubaushene report (1).

RESULTS AND DISCUSSION

The survey results for total coliforms and heterotrophic bacteria are presented on maps of the survey area (Figure 1 - 5). All data presented on the map are the geometric means (GM) for FC, the GM given is per 100 ml while that for HB is per 1 ml.

The FC and FS results are presented in Tables I and II. These are the actual counts that appeared at the station(s) on a given day. All other samples had levels below the detectable limit of 4/100 ml.

The bacterial water quality of the Penetang-Waubaushene survey area in 1976 was generally good. Levels of sanitary indicator bacteria were, with a few exceptions, acceptable for recreational use and public surface water supplies throughout the survey period.

During the May survey (Figure 1), the mean of the TC density was 18/100 ml for the entire area when only stations that had bacterial levels above the detection limit were considered. When the results were adjusted to include areas with non-detectable levels of bacteria, the overall GM was 5 TC/100 ml and two areas (Midland and Penetang STP's) had significantly higher TC densities. The entire area was homogeneous for heterotrophic bacteria and has a GM of 1480 HB/ml with the exception of a higher level at the Midland STP. Fecal coliform and fecal streptococcus results were low with only 3% of all samples above the minimum detectable level. This would indicate that there was little fecal input. However, on April 29, 128 FS/100 ml were found at Midland STP. This coincided with a peak in the HB count and was possibly due to the hydraulic loading problem at Midland STP (2) and late spring runoff.

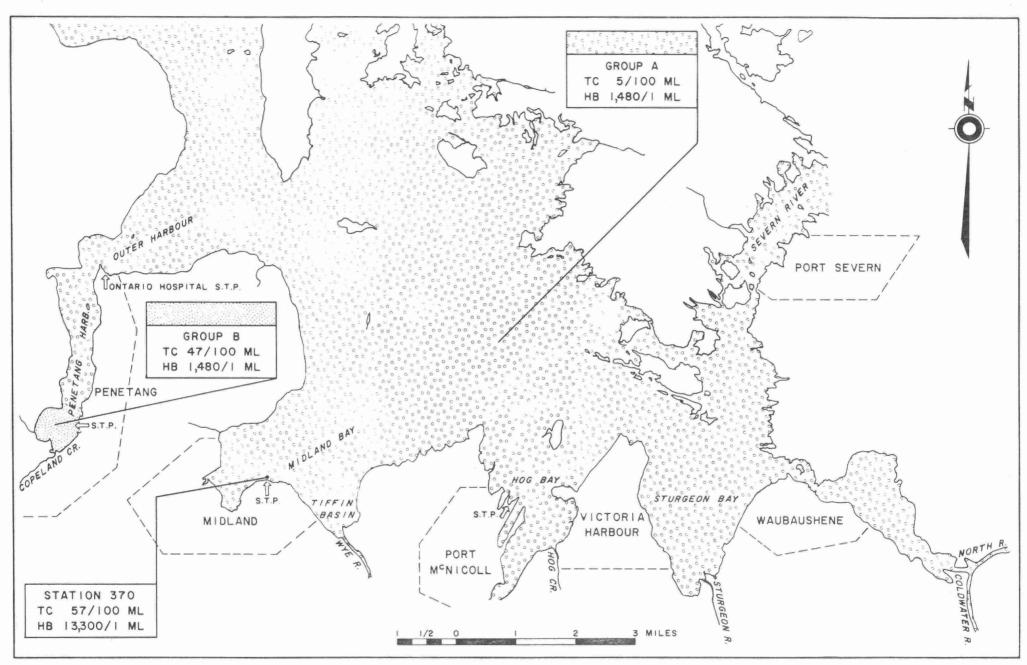


FIGURE 1 - BACTERIAL DISTRIBUTION FOR THE MAY 1976 SURVEY.

DETECTABLE FC LEVELS AT PENETANG-WAUBAUSHENE, 1976

TABLE 1

DATE	STATIONS	FC/100 ML
April 29	371*	20
May 26	354, 367, 370* 369	4 12
June 9	369	4
June 23	353, 367, 369*	4
July 8	369*	4
July 21	363 369* 372	4 16 76
August 4	370*	8
August 18	356 369*	4 12
September 1	355, 358, 359, 360 362, 370*, 372* 363 369*	4 24 48

^{*} indicates STP

TABLE II

DETECTABLE FS LEVELS AT PENETANG-WAUBAUSHENE, 1976

DATE	STATIONS	FS/100 ML
April 29	360, 362, 371* 370*	4
May 13	355, 358	4
June 9	355, 357, 366, 369*, 370*, 371* 368	4 12
June 23	355, 366, 372* 371* 365	4 8 16
July 8	355, 370* 371	4 20
July 21	353, 355, 369*	4 8
September 1	354, 363 362	4 8

^{*} indicates STP

A homogeneous group was formed by the total coliforms during both the June (Figure 2) and July (Figure 3) surveys. Although the GM increased (26 TC/100 ml in June to 37 TC/100 ml in July), the level remainded well within all criteria. When the non-detectable levels of TC were taken into account, the geographic groupings did not change but the GM was 14/100 ml in June and 22/100 ml in July. Very low levels of P. aer. (2/100 ml) were isolated at station 364 and the Penetang STP on July 21. This was the only time throughout the sampling season that P. aer. was isolated. The June HB levels were higher in Penetang Bay (Stations 354, 369) and at the Midland STP while in July, three open water sampling locations (Stations 356, 357 and 365) had significantly lower counts than the rest of the survey area.

The TC concentration in August was 22/100 ml for the whole survey area (Figure 4) but, when the data below the detectable limit was included in the calculations, the GM was 11 TC/100 and the Midland STP had a significantly higher level of 118 TC/100 ml. The heterotrophic density was homogeneous and the GM of the survey area was 1700 HB/ml. Days with elevated FC and FS counts were preceded by a day of fairly heavy precipitation.

The total coliform concentration during the final survey in September was 5 TC/100 ml with the exception of lower Penetang Bay, Midland STP and Sturgeon Bay which were significantly higher (Figure 5). HB data was only available for one day so the analyis of variance could not be performed. Locations with counts well above the overall GM were arbitrarily grouped according to geographic proximity and a new group mean was calculated. Elevated FC levels (Table I) again coincided with rain on the previous day.

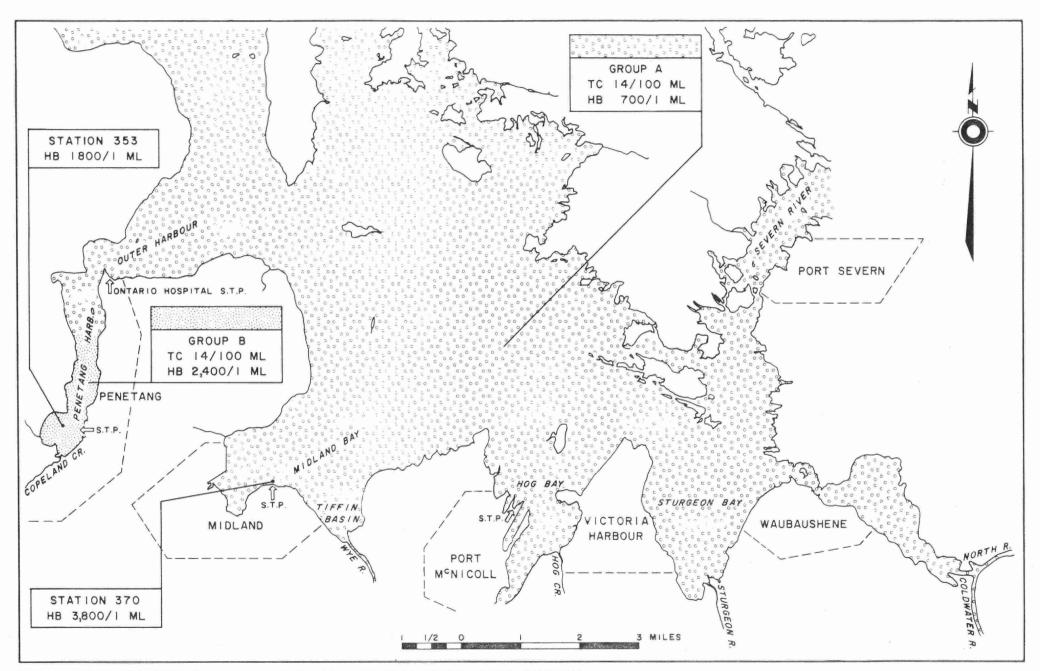


FIGURE 2 - BACTERIAL DISTRIBUTION FOR THE JUNE 1976 SURVEY.

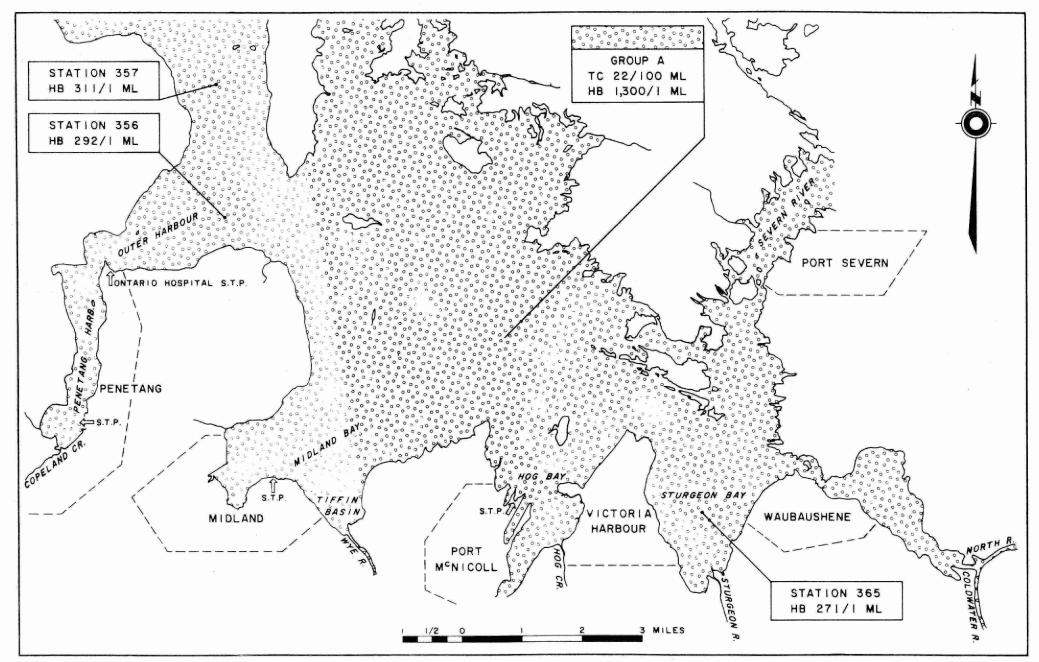


FIGURE 3 - BACTERIAL DISTRIBUTION FOR THE JULY 1976 SURVEY.

The total coliform levels tended to rise to a peak in July then decreased in the 1973, 1974 and 1976 surveys. However, there appears to have been some overall improvement in water quality between 1973 and 1976. In 1976, the overall geometric means for TC and FC were within the Private Water Supplies Criteria requiring chlorination only, whereas in 1973 and 1974, the Recreational Use Criteria was exceeded a number of times.

A statistical comparison of 1974 and 1976 HB concentrations is of limited value since in 1976 the medium was changed from Foot and Taylor to CPS agar and testing has shown that recoveries on CPS are higher (3). However, the HB levels did show a small decrease between 1974 and 1976, and this could indicate a reduction in the nutrient enrichment of the survey area.

The effluents from the Penetang and Midland STP's probably exert some influence on the survey area as these were the locations where any elevated bacterial levels were usually found. Bacterial concentrations in these locations still complied with the Permissible Criteria for private water supplies requiring chlorination and filtration. In 1973 and 1974, Penetang STP appeared to have the greatest effect on water quality. By 1976, the TC and HB counts tended to be higher at Midland STP. There was an overall improvement, however, in the Midland effluent between 1973 and 1976. The expansion and improvement carried out at the waste treatment facilities in the area would appear to be having a beneficial effect on the water quality of the survey area. This is important since this is a major summer recreation region.

The results of these surveys also indicated that areas with bacterial concentrations below the detection limit should be taken into account during the analysis of the data. The value used in these cases in a "less than" value (e.g. less than 20 TC/100 ml) and the resulting GM is not a statistically valid number. The concentration indicated is still higher than the actual concentration, however, it is closer to the true GM than when the areas with non-detectable levels are ignored. In addition, their inclusion results in the data being more sensitive to the geographic heterogeneity of the bacterial levels.

SUMMARY AND CONCLUSIONS

The 1976 surveys indicated that the bacterial quality of the water in the Penetanguishene-Waubaushene area had improved since 1973-74.

The effect of the Penetang and Midland STP's was still evident, but improvement of waste treatment facilities has had a beneficial effect since 1973-74. The whole area was acceptable throughout the 1976 survey period for recreational use and as a private water supply subject to chlorination and filtration. In all cases, use of any surface water without treatment is never recommended.

Periodic monitoring of the area should be carried out in future to ensure that the water quality does not begin to deteriorate. Surveys of the intensive type are recommended since these are less likely to reflect weekly periodic bacterial fluctuations than biweekly surveys which are generally taken on the same day of the week. Intensive surveys also tend to be more sensitive to geographical heterogeneity (1).

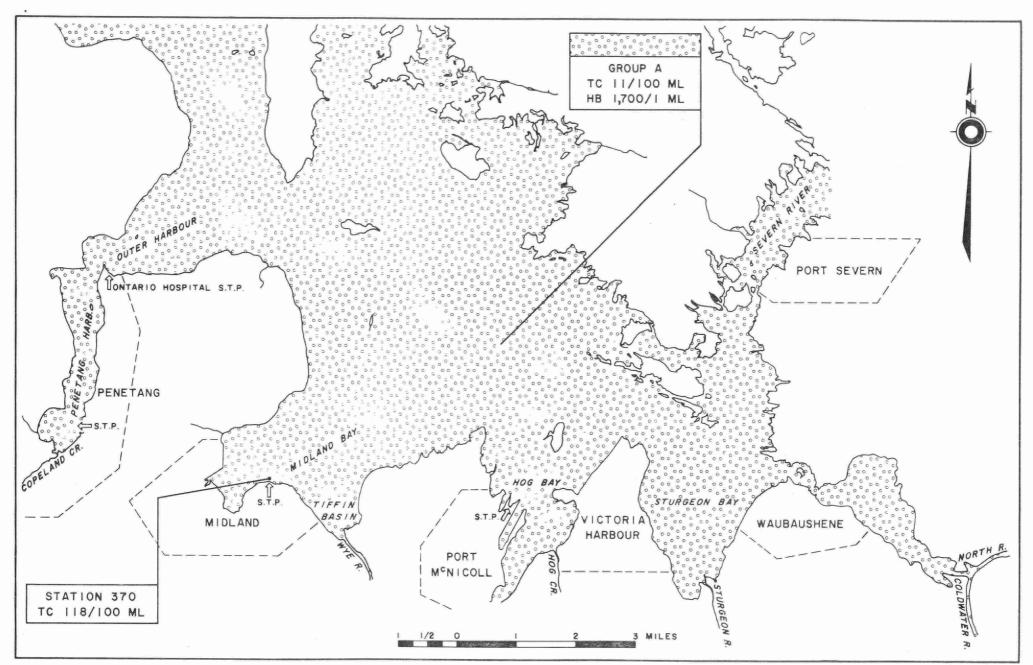


FIGURE 4 - BACTERIAL DISTRIBUTION FOR THE AUGUST 1976 SURVEY.

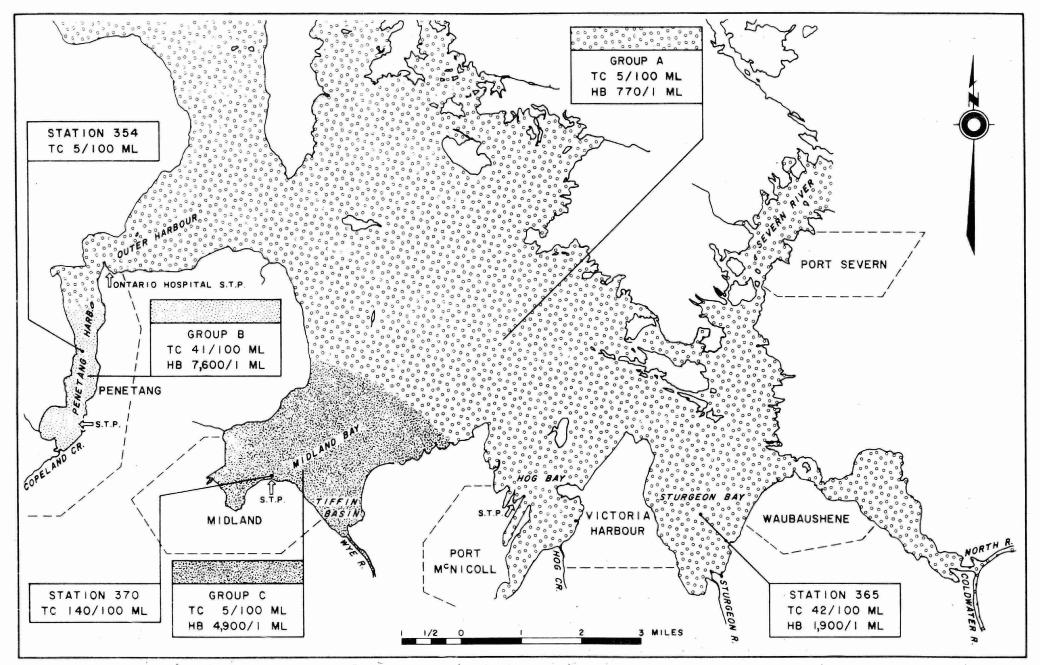


FIGURE 5 - BACTERIAL DISTRIBUTION FOR THE SEPTEMBER 1976 SURVEY.

REFERENCES

- Young, M. 1977. A Bacteriological Study of the Penetanguishene-Waubaushene Area of Georgian Bay 1973-74. Ministry of the Environment, Laboratory Report.
- 2. Standard Methods for the Enumeration of Water and Wastewater, 13th edition, 1971. APHA, AWWA, WPCF.
- Young, M. 1977. Comparison of four plating techniques and four media for the enumeration of heterotrophic bacteria. Ministry of the Environment, Laboratory Report.
- 4. Levin, M.A. and V.J. Cabelli. 1972. Membrane filter techniques for enumeration of Pseudomonas aeruginosa. Appl. Microbiol. 24, 864-870.
- 5. Marton, F. Personal Communication, Ministry of the Environment, Central Region, 1975.

